Background

- *Plasmodium falciparum* is the most common human malaria parasite species in Sub-Saharan Africa, where the continent bears over 90% of the global *P. falciparum* burden.
- In the past decade, Ghana has made remarkable progress in malaria control. The percentage of outpatient attendance in public health facilities decreased sharply from 48% to 28% from 2008 to 2016.
- However, in some parts of the country, malaria still remains one of the leading cause of morbidity and mortality. For instance, previous studies showed a wide variation in parasite prevalence between the south (4%) and the north (51%)².
- Seasonal Malaria Chemoprevention (SMC) is one of the preventive measures aims to protect infants and young children in highly endemic areas with seasonal transmission.
- The SMC intervention has a potential to result in the emergence of antimalarial drug resistant parasites³. An accurate profile of genetic structure and diversity of the parasites is essential to infer where drug resistance genotypes originated and how fast they spread⁴.

Objectives

- This study aims to genotype the parasite populations from different transmission zones using microsatellites and infer genetic diversity, population structure and underlying transmission patterns.
- Our goal is to generate baseline data that allows us to assess how current interventions influence the transmission dynamics and complexity of infections in the asymptomatic populations.

Methods

- Asymptomatic samples were collected from five sites representing the three distinct ecological zones including the northern savannah, the central forest, and the southern coastal regions.
- Fingerprick dried blood samples were collected from over 1,000 schoolchildren aged 3-12 during the rainy season (July-August) of 2017.
- Parasite DNA was screened by quantitative real-time PCR based on 18S rRNA and TARE-2 gene regions⁵.
- Six microsatellite loci with tri- or tetranucleotide repeats were typed for *P. falciparum* using the published oligonucleotide primers⁶.
- Gene codons in pfcrt (K76T) and pfmdr1 (N86Y and Y184F) associate with amodiaquine (AQ) resistance; *pfhr1* (C511, C59R, S108N) and *pfhr1* (A437G and K540E) associate with sulfadoxine-pyrimethamine (SP) resistance were examined by sequencing.

Results

**Figure 2. Within-host diversity by microsatellites**

- Only samples of a single or bi-clonal infection were included. The majority of samples in the north contain more than two clones and thus were not included in the analyses.
- Six samples from the south (marked in asterisk) have clones that are genetically different from each other. The rest of the samples mostly from the central region have clones that are genetically similar.
- Ongoing work use *pfmsp1* to further infer multiplicity and diversity of polyclonal samples.

**Figure 3. Genetic relatedness of *P. falciparum* samples from different transmission zones.**

- Each individual contain either a single or multiple parasite strains.
- Phylogenies was constructed based on the presence or absence of an allele and does not reflect the genetic identity of individual clones.
- Parasites in the north and central are more closely-related compared to those in the south.

**Figure 4. Bayesian barplot showing genotyping structure among *P. falciparum* samples.**

- Two most probable genetic clusters (pink and blue) were identified based on Bayesian inference.
- Parasites in the north and central populations shared predominantly the blue cluster, whereas parasites in the south had the pink cluster.

**Figure 5. Resistance marker prevalence in Ghana.**

- Less than 5% of the samples harbor the mutant genotype at pfcrt codon 76 and pfmdr1 codon 86.
- Over 90% of the samples carried a triple mutant *pfhr1* genotype (108N, 511, and 59R). A high prevalence of *pfhr1* 108N mutation suggested strong resistance to SP.
- Nearly 90% of the samples showed *pfhr1* mutation at codon 437 but not 540.

Summary

- A higher rate of polyclonality was observed in the north compared to the south and central regions.
- Parasites in the northern and central populations are genetically closely related, possibly due to frequent gene flow.
- Prevalence of the resistant genotypes were also similar between the north and central populations.
- A large percentage of samples observed with *pfhr1* and *pfhr1* mutations, suggestive of increased resistance could be due to selective pressure by the SMC implemented since 2015.
- By contrast, a few samples had *pfhr1* mutation associated with CQ resistance.

References


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